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THE NIACIN CONTENT OF ALBERTA GRAINS

by

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UNIVERSITY OF ALBERTA

April 1947

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THE NIACIN CONTENT OF ALBERTA GRAINS

Harold Daniel Simonson
Department of Animal Science

A THESIS
submitted to the University of Alberta
in partial fulfillment of the
requirements for the degree of
MASTER OF SCIENCE

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THE NIACIN CONTENT OF ALBERTA GRAINS

INTRODUCTION

The therapeutic value of niacin in the treatment of pellagra as reported by Spies and associates (1) has directed considerable attention to the possible role that this vitamin may play in animal and poultry nutrition. Pathological changes in animals maintained on rations low in niacin are being studied in an attempt to associate niacin deficiency symptoms with recognized diseases appearing in the field.

A knowledge of the niacin content of the feeds being used is essential to animal experimentation. In Canada evaluation of the niacin content of rations has been based upon values reported from researches conducted mainly in American Institutions on relatively few samples of feeds. These feeds were grown under quite different environmental conditions from those existing in Western Canada. In order to determine the niacin content of cereal grains grown in Alberta, together with any relationship that may exist between the niacin and protein content of these grains, a survey was undertaken at the University of Alberta in the fall of 1945.

The grain samples studied in this survey were obtained from two sources:

(a) crop samples collected for another project entitled "Soil and Plant Relations Affecting the Nutritional Value of Foods and Feeds".

(b) hybrid wheats grown at Fallis and Edmonton during the years 1945 and 1946.

While these samples were not collected expressly for niacin assay, they afforded adequate material for a study of the distribution of this vitamin in wheat, oats and barley grown on a wide variety of soils.

The results of this study, preceded by a brief review of animal and poultry investigations, are presented in this thesis.

ANIMAL AND POULTRY EXPERIMENTATION

In 1937 Birch, Chick and Martin (2) carried out trials with pigs maintained on pellagra-producing diets composed chiefly of maize. During the course of their experiments a disorder arose which they concluded was due to a lack of some constituent other than protein. Substitution of a barley and wheat mixture for the maize provided sufficient of the unknown constituent to promote normal growth. The following year Chick, Macrae, Martin and Martin (3) reported the curative action of niacin when added to a hog ration composed of white maize, pea meal, purified casein, cod liver oil and salt mixture.

The first report of a nutritional disorder simulating hog pellagra actually occurring in a commercial herd of swine was that of Madison, Miller and Keith in 1938 (4). The herd had been maintained on a ration composed of corn, oats,wheat middlings and a limited amount of skim milk. The death loss was considerable and the remaining pigs showed symptoms of anorexia, diarrhea and stunted growth. A scurf appeared on the ears and body. The addition of 50 milligrams of niacin to the ration of each pig per day brought about complete recovery within a few weeks.

Davis, Freeman and Madsen, as quoted in (5), noted the appearance of necrotic enteritis in swine and observed that the incidence of the disease was more pronounced as the proportion of corn in relation to other grains in the ration was increased. It was concluded that the higher incidence was due to a niacin deficiency.

In commenting on the results of these earlier experiments, Wintrobe and collaborators (6) indicate that the diets were probably incomplete in several respects, and that the observed symptoms of malnutrition were not due to niacin deficiency alone. Recently they showed (7) that excellent growth and development were obtained when supplements of pyridoxine, choline and calcium pantothenate were fed in addition to thiamine, riboflavin and niacin. They then conducted a trial in which niacin was omitted and no ill effects were observed. In trying to give some plausible explanation to their observations, they concluded that the high protein

content of the diets used might have been a factor of importance and proceeded to test the validity of this theory. They found that pigs fed a high protein diet (26.1% casein) supplemented with choline, pyridoxine, calcium pantothenate, thiamine and riboflavin, but not with niacin, showed no signs of nutritional deficiency except for retarded growth in certain instances. When the protein content of the diet was low (10% casein) the omission of the niacin was associated with impaired growth, rough coats, diarrhea, poor appetite and anemia. There seems to be a close relationship between protein and niacin requirement.

Recognition of the importance of niacin in poultry feeding is even more recent. Briggs, Mills, Elvehjem and Hart (8) demonstrated that chickens require a dietary source of niacin when fed certain purified diets. Briggs, Groschke and Lillie (9) showed that the amount of niacin required by chickens was associated with the protein content of the ration, and indicated that protein quality as well as quantity is an important factor. The possibility that turkey poults may suffer from a niacin deficiency was also studied by Briggs (10). He found that a deficiency in poults was associated with poor growth, inflammation of the mouth, diarrhea and perosis. He concluded that for normal growth turkey poults require two to three times as much niacin as do Leghorn chicks.

While this review deals only very briefly with some

of the work that is being carried out to determine the significance of niacin in poultry and animal feeding, it indicates that considerable emphasis is being placed on this phase of nutrition.

Niacin Requirements

Data on the niacin requirements of farm animals are few and from the preceding discussion it is apparent that the requirement is linked with the quality of the ration. Dietary requirements that have been suggested for a number of species on the basis of results obtained with purified diets are listed in Table I.

TABLE I

Dietary Niacin Requirement of Some Farm Species

Turkey poults	3 - 5 mg./100 grams feed (10)
Chicks	1.5- 2 mg./100 grams feed (11)
Pigs	5 -10 mg./100 lbs. live weight (12)
Dogs	200 - 225 μg/kg. body weight per day (13)
Ruminants	Not required (14)

Niacin Content of Cereal Products

Information on the content and distribution of niacin in cereal products is rather limited. The values reported have been obtained using either chemical or microbiological procedures (15-21). The report of Brown, Thomas and Bina (16) in 1946 gives the most recent results obtained by the micro-

biological procedure of Barton-Wright (15). The values given for wheat, oats and barley plus the results on a number of other products are listed in Table II. No statement is made regarding the number of samples of any one grain or by-product assayed.

TABLE II

The Niacin Content of a Number of Cereals and Cereal Products (16)

Material	Niacin µg/gram
Barley	65.0
Wheat	53.0
Oats	7.2
Rye	8.7
Corn	15.7
Rice (milled)	11.8
Wheat bran	214.8
Wheat germ	54.4

The data in Table II suggest that wheat and barley constitute relatively good sources of niacin, while rice, corn, rye and oats are low in this vitamin. Wheat bran is much richer in niacin than is whole wheat or wheat germ which indicates that there is an accumulation of niacin in the bran portion of the wheat kernel. These values represent the total niacin content of the cereals and do not differentiate between the relative amounts of niacin found as the free acid, free niacin amide and niacin amide associated with coenzymes I and II. This phase of the subject has been investigated recently by Krehl and coworkers (22) who have presented data in which

the free niacin amide and the amide associated with coenzymes I and II are given as percentages of the total niacin. The amide was found to be concentrated in the germ while the bran portion of the kernel was found to be rich in free niacin. It is interesting to note that the total niacin values obtained by Krehl and coworkers (22) by using Leuconostoc mesenteroides check reasonably well with those for similar products reported by Bina, Thomas and Brown (16) using Lactobacillus arabinosus.

PART I

A. MICROBIOLOGICAL ASSAYS FOR NIACIN

Assay materials

Grain samples collected for another project, "Soil and Plant Relations Affecting the Nutritional Value of Foods and Feeds", which was being studied at the University of Alberta, were available and provided excellent material for a survey of the niacin content of Alberta grown cereals. Samples of wheat, oats and barley were collected during the crop years 1944, 1945 and 1946 from the major soil zones of Alberta. Variations in conditions of soil and climate under which the grains were grown are reflected in the wide variations in the protein content of the samples.

Samples were prepared for assay by grinding through the

1 mm. screen of a Wiley mill. Ground samples were stored in glass bottles at room temperature.

Assay Procedure

The assay procedure used was essentially the microbiological method of Snell and Wright (23) and its modifications as described by Krehl *et al*. (24).

Principle

The production of acid by Lactobacillus arabinosus 17-5 in tubes containing known amounts of niacin was measured and plotted against micrograms of niacin. The niacin content of the grain samples was calculated from this standard curve.

Reagents

Stock solutions were stored in the refrigerator at a temperature of approximately 4°C. A thin layer of toluene and a few drops of chloroform were added as preservatives. The solutions and reagents used, together with the method of preparation, are listed below.

(a) Acid hydrolyzed casein solution was obtained from General Biochemicals, Incorporated. Each ml. was equivalent to 100 mg. of casein.

(b) 1 - Tryptophane - one gram of 1-tryptophane was dissolved in about 300 ml. of warm water, with the aid of a few drops

of concentrated hydrochloric acid. The solution was diluted to 500 ml. so each ml. contained 2 mg. of l-tryptophane.

(c) l-Cystine - two grams of l-cystine were suspended in 300 ml. of hot water and a small amount of concentrated hydrochloric acid was added to facilitate solution. The solution was diluted to 500 ml. so that each ml. contained 4 mg. of l-cystine.

(d) Anhydrous C.P. glucose and sodium acetate were weighed out as required.

(e) Calcium pantothenate, thiamine chloride and pyridoxine solution - prepared in distilled water in such a manner that each ml. contained 100 micrograms of each of these vitamins.

(f) Riboflavin - a solution containing 100 micrograms of riboflavin was prepared in 0.02 N acetic acid. Unnecessary exposure to light was avoided and a fresh solution was prepared at frequent intervals.

(g) Biotin - the contents of an ampule containing 25 micrograms of biotin as the free acid were diluted to 250 ml. so that each ml. contained 0.1 microgram of biotin.

(h) P-aminobenzoic acid - fifty milligrams of p-aminobenzoic acid were dissolved in about 300 ml. of distilled water and diluted to 500 ml. so that each ml. contained 100 micrograms of p-aminobenzoic acid.

(i) Adenine, guanine and uracil - a solution was prepared to contain 1 mg. per ml. of each of these constituents.

Solution was effected by prolonged heating in the presence of a few drops of hydrochloric acid. This solution was renewed at frequent intervals.

(j) Inorganic salts, solution A - 25 g. of K_2HPO_4 and 25 g. of KH_2PO_4 were dissolved in water and made up to 250 milliliters.

(k) Inorganic salts, solution B - 10 g. of $MgSO_4.7H_2O$, 0.5 g. of NaCl, 0.5 g. of $FeSO_4.7H_2O$ and 0.5 g. of $MnSO_4.4H_2O$ were dissolved in water and diluted to 250 milliliters.

(l) Niacin solutions - five hundred mg. of niacin were dissolved in about 300 ml. of warm distilled water and diluted to 500 milliliters. This stock solution was made up at least once every two months. Exactly 10 ml. of the stock solution were diluted to 1000 ml. to give a concentration of 10 micrograms per milliliter. A 10 ml. aliquot of this dilution was then diluted to 1000 ml. to make the standard solution containing 0.1 micrograms of niacin per milliliter. The last two dilutions were made each time a niacin determination was run.

Assay Medium

The amounts of stock solutions and reagents used in preparing 500 ml. of the assay medium are given in Table III.

TABLE III

Composition of the Assay Medium

Acid hydrolyzed casein	50 ml.
l-cystine solution	50 ml.
l-tryptophane solution	50 ml.
Anhydrous glucose	20 grams
Anhydrous sodium acetate	20 grams
Biotin solution	2 ml.
p-amino benzoic acid solution	1 ml.
Thiamine, calcium pantothenate and pyridoxine solution	1 ml.
Riboflavin solution	2 ml.
Adenine, guanine and uracil solution	10 ml.
Inorganic salts, solution A	5 ml.
Inorganic salts, solution B	5 ml.

After mixing the medium was adjusted potentiometrically to pH 6.8 using 6 N sodium hydroxide solution and the volume was made up to 500 ml. with distilled water. The medium was freshly prepared each time a determination was carried out.

Stock Cultures and Inoculum

The organism employed was Lactobacillus arabinosus 17-5 obtained from the American Type Culture Collection, Georgetown University, Washington, D. C. Stock cultures of the organism were carried in yeast extract dextrose agar composed of 0.5% dextrose, 1% yeast extract, 0.5% sodium acetate and 1.5% agar. The cultures were preserved in a refrigerator at a temperature of approximately 4°C. and were renewed at weekly intervals.

To grow the inoculum for the assay tubes a transfer was made from one of the stab cultures into a tube containing 5 ml.

of the medium shown in Table III plus 5 ml. of distilled water and one microgram of niacin. This liquid culture was incubated for 24 hours at 37°C. and the cell suspension so obtained was packed by centrifugation. The liquid was poured off and the cells were resuspended in 10 ml. of 0.9 percent saline. This solution constituted the inoculum.

Preparation of extracts

In preparing the extracts for assay the initial weight of sample used depended upon the niacin content of the grain. Two grams of barley, wheat and oats constituted a sample of satisfactory size. The weighed samples were transferred to Erlenmeyers and 40 ml. of 1 N hydrochloric acid were added. The contents were mixed and autoclaved for twenty minutes at 15 pounds pressure. Filtration through coarse filter paper following autoclaving removed the coarse grain particles and facilitated subsequent pipetting. The filters were washed three or four times with distilled water and the washings were added to the filtrate. The filtrates were made up to 200 ml. in volumetric flasks. The flasks were shaken well and 10 ml. aliquots of the wheat and barley filtrates and 20 ml. aliquots of the oat filtrates were diluted to about 30 ml. with distilled water. The pH of each was adjusted potentiometrically to 6.8 after which the wheat and barley solutions were made up to 200 ml. and the oat solution to 100 ml. in volumetric flasks. The extracts at this stage were ready for tubing.

Preparation of assay tubes

It was found that the greatest efficiency was obtained by analyzing sufficient samples each time to warrant the use of 100 assay tubes and 500 ml. of the medium previously described. Five ml. of the medium were transferred to each tube. Eight pairs of tubes were prepared for the blanks and standard niacin curve. To the series of tubes were added 5 ml. of water (blank), 0.5, 1.0, 1.5, 2.0, 2.5, 3.0 and 3.5 ml. of standard niacin solution (0.1 μg/ml.). In each case sufficient distilled water was added to bring the final volume of each tube to 10 milliliters. The remaining tubes were used for the assay of the test material. In the original analyses each assay was carried out at five different levels, set up in duplicate, using 1.0, 2.0, 3.0, 4.0 and 5.0 ml. of extract. After considerable experience only the 2.0, 3.0 and 4.0 ml. levels of assay were used. The volume of liquid in each tube was brought to 10 ml. by the addition of distilled water. The tubes were plugged with cotton, autoclaved at 15 pounds pressure for fifteen minutes, cooled to room temperature and inoculated with one drop of inoculum. After 72 hours incubation at 37°C. the contents of each tube were titrated to pH 6.8 with 0.1 N sodium hydroxide solution using a Beckmann Potentiometer with outside electrodes. The standard curve, a typical example of which is illustrated in Figure 1, was plotted and from it the niacin content of each assay tube was determined. The final niacin content of each sample was based on the average

values for the different levels of assay.

Validity of Procedure

The procedure described has been used extensively by other workers in this field and possible sources of error have been reduced to a minimum. Typical results obtained in separate assays on the same sample are shown in Table IV. It may be mentioned that much closer checks were normally obtained on wheat and oats than on barley.

TABLE IV

Reproducibility of Results

Cereal	Niacin Content µg/g.	
	Assay 1	Assay 2
Wheat		
1	63	63
2	53	55
3	63	65
4	65	69
Oats		
1	20	19
2	11	12
3	8	8
4	14	13
Barley		
1	69	71
2	70	71
3	67	74

The results of assays run to determine the recovery of known amounts of niacin added to samples prior to hydrolysis

are shown in Table V. The data indicate an error of less than ± 10% as measured by recovery of added niacin. The mean for the values shown in Table V under "per cent recovery" is 97.4 per cent.

TABLE V

Niacin Recovery in Grain Samples

Sple.	Sple. + 25 µg. niacin	Sple. without added niacin	Recovery by difference	Per cent recovery
W - 1	85.8	62.3	23.5	94.0
W - 2	78.8	52.5	26.3	105.2
W - 3	77.7	53.2	24.5	98.0
B - 1	89.5	66.7	22.8	91.2
B - 2	100.8	76.8	24.0	96.0
O - 1	45.0	19.7	25.3	101.2
O - 2	34.9	10.8	24.1	96.4

A few samples of finely ground grain were reassayed after eight months' storage at room temperature in brown glass bottles. The results of the initial and final assays, as shown in Table VI, indicate that niacin is stable in ground grain stored under these conditions.

TABLE VI

The Effect of Storage on the Niacin Content of Ground Cereal Grains

Cereal	Niacin µg/g. Initial Assay	Niacin µg/g. Eight Months later
Wheat		
1	69	67
2	52	47
Oats		
1	20	20
2	12	11
Barley		
1	63	67
2	77	77

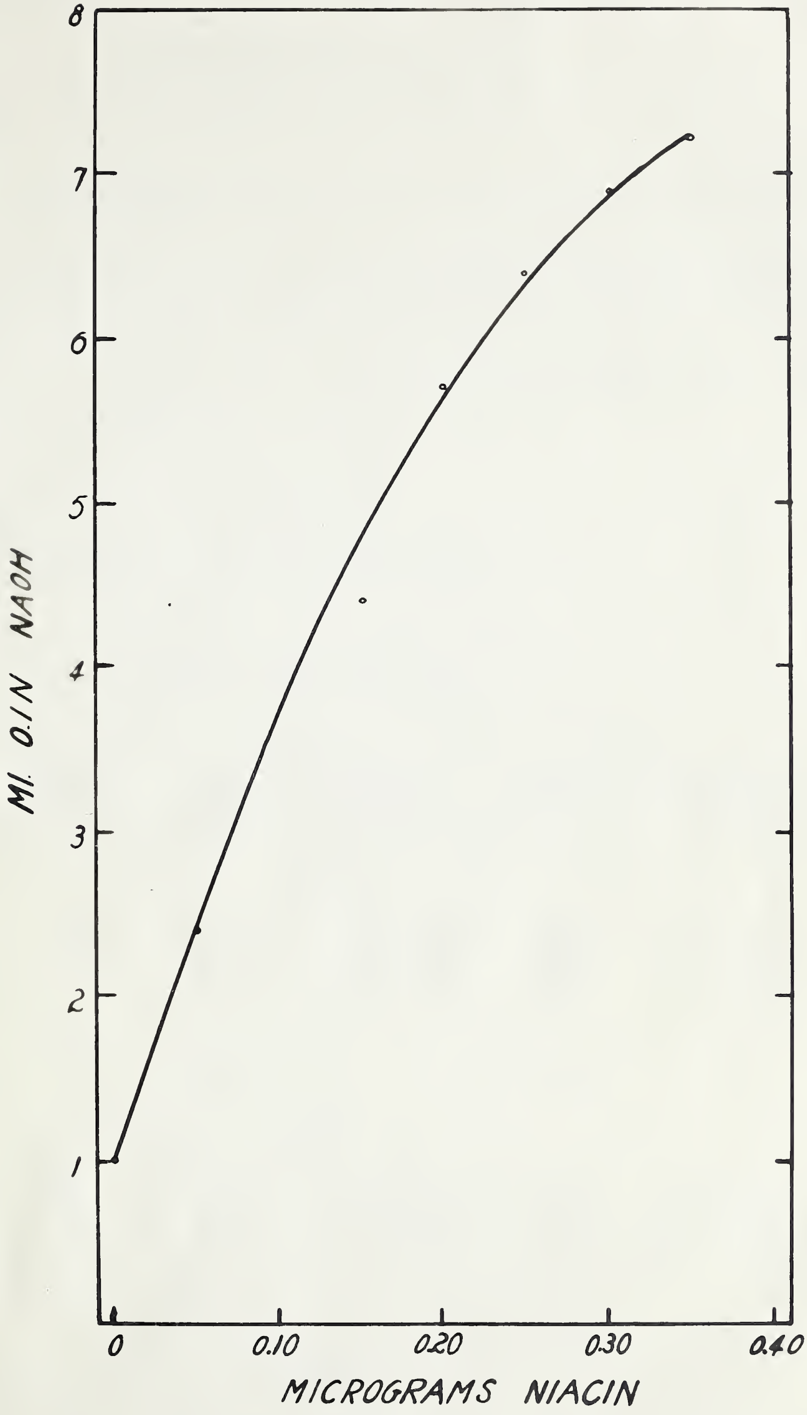

Fig.1. STANDARD NIACIN CURVE

RESULTS AND DISCUSSION

The initial assays were carried out on the 1944, 1945 and 1946 wheat, oat and barley samples obtained from the various soil zones of the province. The summarized data are presented in Table VII. The individual assay values are compiled in Appendix Tables I - IX.

TABLE VII

Niacin and Protein Results by Years
for Wheat, Oats and Barley.
1944, 1945 and 1946.

Year	No. of Samples	Niacin		Protein	
		Mean µg/g.	Range µg/g.	Mean %	Range %
		Wheat			
1944	24	49.8	35.3-74.0	12.9	8.6-17.3
1945	23	59.6	45.7-72.3	14.0	8.7-19.7
1946	8	59.9	43.4-77.9	13.0	8.7-16.9
Total	55				
Mean		55.4		13.4	
		Oats			
1944	27	8.7	7.0-14.3	11.0	6.6-15.9
1945	28	12.1	8.0-18.7	12.7	8.1-16.9
1946	8	8.8	7.3-11.6	12.6	10.3-14.9
Total	63				
Mean		10.2		12.0	
		Barley			
1944	28	69.8	49.3-93.3	12.4	8.3-16.2
1945	24	71.2	58.7-87.3	13.8	9.4-17.1
1946	8	72.0	59.7-85.4	13.3	9.4-15.9
Total	60				
Mean		70.7		13.1	

The data in Table VII indicate that, of the three cereal grains, oats has the least, wheat an intermediate and barley the greatest amount of niacin with means of 10.2, 55.4 and 70.7 micrograms per gram respectively. Oats, as a cereal, represents a poor source of niacin whereas barley and wheat are relatively good sources of the vitamin. Results published by other workers for grains grown in other regions substantiate these findings (16, 18). Very few data on the niacin content of Canadian grains have been published. Barton-Wright (15) assayed a few Manitoba wheat samples, the mean niacin content of which was 63.3 micrograms per gram. Similar data pertaining to the niacin content of Canadian-grown barley and oats are not available. Data reported in this study indicate extreme variability for any one cereal within any one year. The variability between years as indicated by the mean niacin content for each year, is less for barley than it is for wheat and oats.

The majority of wheat, oat and barley samples assayed were of the varieties Marquis, Victory and Newal. In an attempt to determine whether there were any variations within variety from one soil zone to another and between the years 1944 and 1945, the data were grouped as follows: (1) samples from the gray and gray transitional, (2) the black, shallow black and black transitional and (3) the brown and dark brown zones. The summarized data are presented in Table VIII. The individual assay results used in compiling this table, and miscellaneous values on samples that did not conform to the main variety represented are given in Appendix Tables X - XIII.

TABLE VIII

Summary of the Niacin Content of 1944 and 1945 Wheat, Oat and Barley Samples,

Grouped According to Soil Zone.

Soil Zone*	Marquis Wheat				Victory Oats				Newal Barley			
	No. of Samples	Mean Protein %	Niacin µg/g.		No. of Samples	Mean Protein %	Niacin µg/g.		No. of Samples	Mean Protein %	Niacin µg/g.	
			Mean	St. Dev.			Mean	St. Dev.			Mean	St. Dev.
G.& G.T.												
1944	7	10.9	46.9±	3.6	9	9.4	8.4±	2.5	6	11.0	71.5±	10.1
1945	2	12.0	54.3±	7.2	10	11.5	12.0±	2.4	7	12.1	72.9±	6.9
B., S.B. & B.T.												
1944	6	13.8	59.0±	7.5	9	11.7	8.4±	1.0	10	13.0	73.5±	11.6
1945	3	14.5	54.2±	5.6	4	12.2	12.6±	2.1	3	14.9	71.4±	7.8
Br.& D.B.												
1944	7	14.2	47.6±	5.1	8	11.8	9.5±	1.9	10	12.5	65.3±	11.0
1945	9	14.1	59.5±	6.1	10	13.8	11.7±	2.8	12	15.0	70.0±	8.4

* G. = Gray; G.T. = Gray transitional; B. = Black; B.T. = Black transitional;
S.B. = Shallow black; D.B. = Dark brown; Br. = Brown.

It is apparent that the niacin content of one variety of a cereal grown in any one soil zone is extremely variable. This variability is particularly noticeable in the barley samples assayed. The niacin content of Newal barley grown in the black soil zone during 1944 ranged from 63.0 to 93.3 micrograms per gram. The average values show some variation in the niacin content of cereal grains from soil zone to soil zone and from year to year; but, in view of the wide variation within soil zones for a single year, it may be difficult, even with **many** more data, to demonstrate that a real difference, attributable to either soil zone or year, exists.

To determine whether or not the niacin content of any one cereal was related to protein content, correlation coefficients between niacin and protein were determined for the years 1944, 1945 and 1946. The factors used in converting the protein to a 13.5 per cent moisture basis were 5.41 for barley and oats and 4.93 for wheat. All niacin assay values, regardless of variety within one cereal, were employed. The coefficients obtained are shown in Table IX. Scatter diagrams representing the 1944 and 1945 relationships between niacin and protein for wheat, oats and barley are shown in figures 2, 3 and 4.

TABLE IX

Correlation Coefficients Between Niacin and Protein Content of Wheat, Oats and Barley Samples in 1944, 1945 and 1946.

Cereal	Number of Samples	Correlation Coefficient	1%	5%
		1944		
Wheat	24	+0.305	0.515	0.404
Oats	27	+0.303	0.487	0.381
Barley	28	+0.066	0.478	0.374
		1945		
Wheat	23	+0.514*	0.526	0.413
Oats	28	+0.545**	0.478	0.374
Barley	24	-0.391	0.515	0.404
		1946		
Wheat	8	+0.879**	0.834	0.707
Oats	8	+0.780*		
Barley	8	-0.206		

The data for the 1944 samples of wheat, oats and barley indicate that no significant relationship exists between niacin and protein content for this year. However, similar data for the 1945 samples indicate significant and highly significant positive relationships for wheat and oats respectively, and a non-significant negative correlation for barley. It is of interest to note that the few data obtained on the wheat and oat samples grown in 1946 indicate trends similar to those observed in 1945. While the negative correlation of -0.206 for barley is not significant, it, along with the trend noted in the 1945

barley samples, suggests that an inverse relationship may exist between the niacin and protein content of this cereal. It is difficult to account for the relationships observed. It appears that niacin accumulation may be affected by a number of factors such as soil, climate and genetic constitution of the cereal.

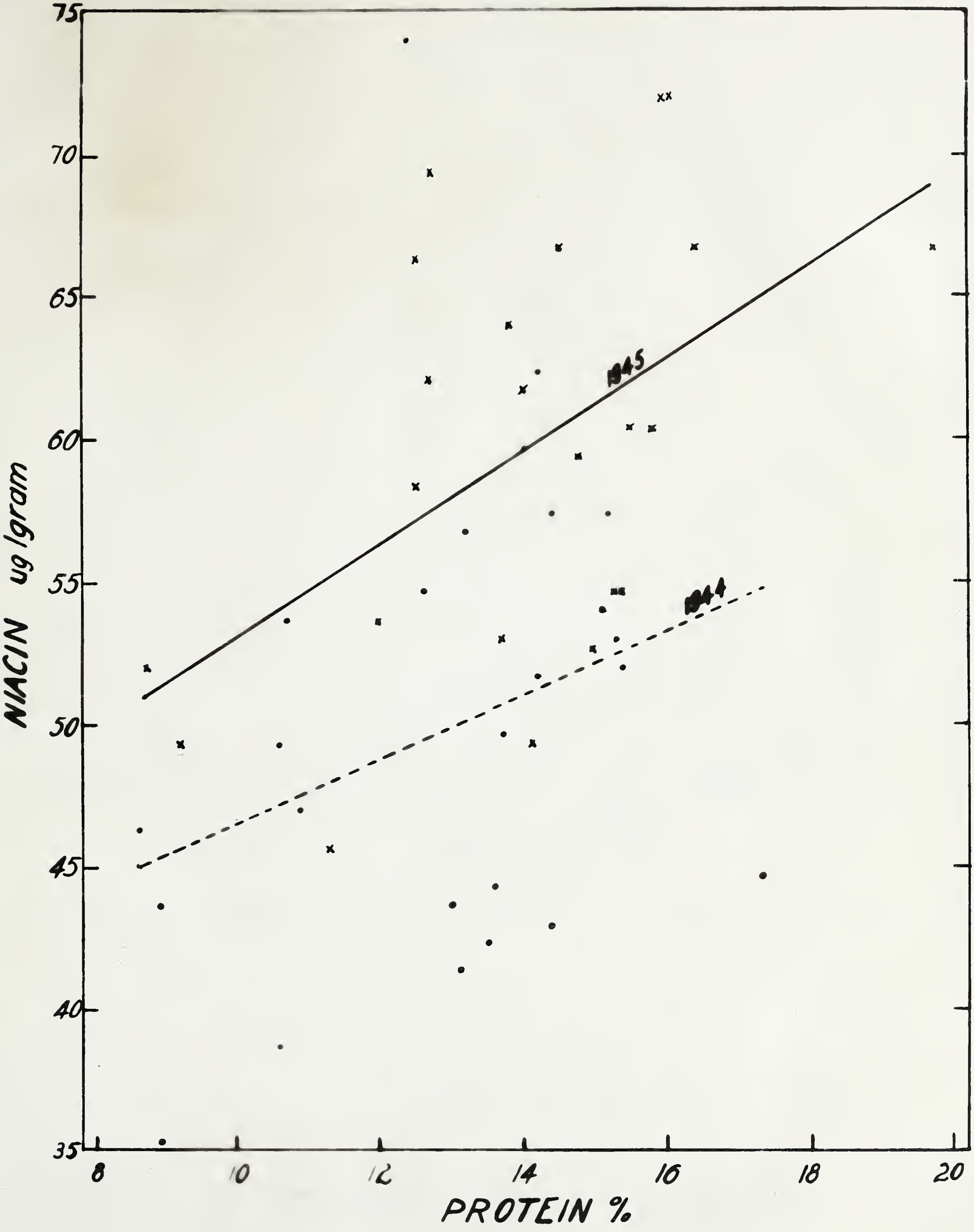

Fig. 2. Relation between niacin and protein content of wheat.
1944 ... 1945 xxx

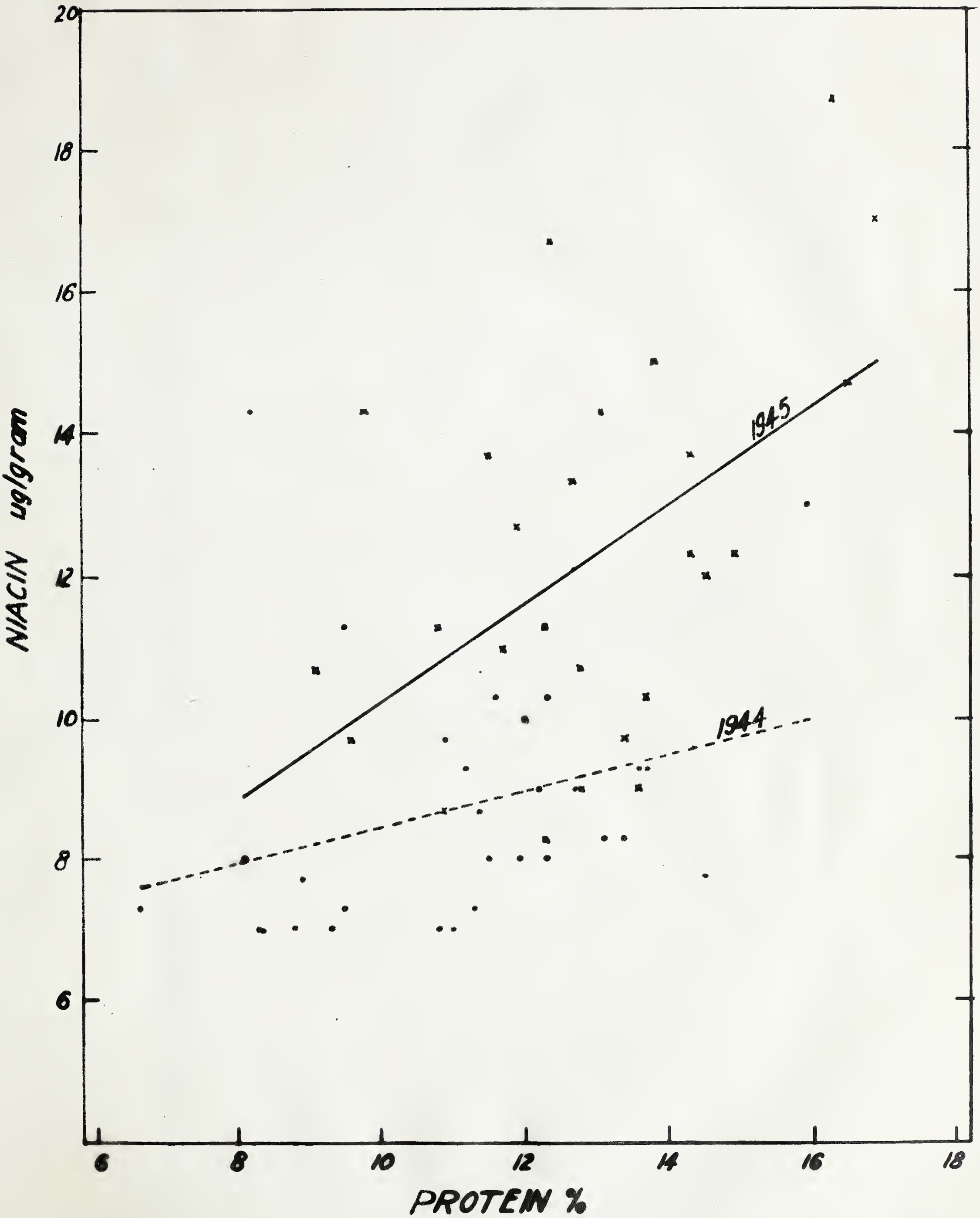

Fig. 3. Relation between niacin and protein content of oats.
1944 ... 1945 xxx

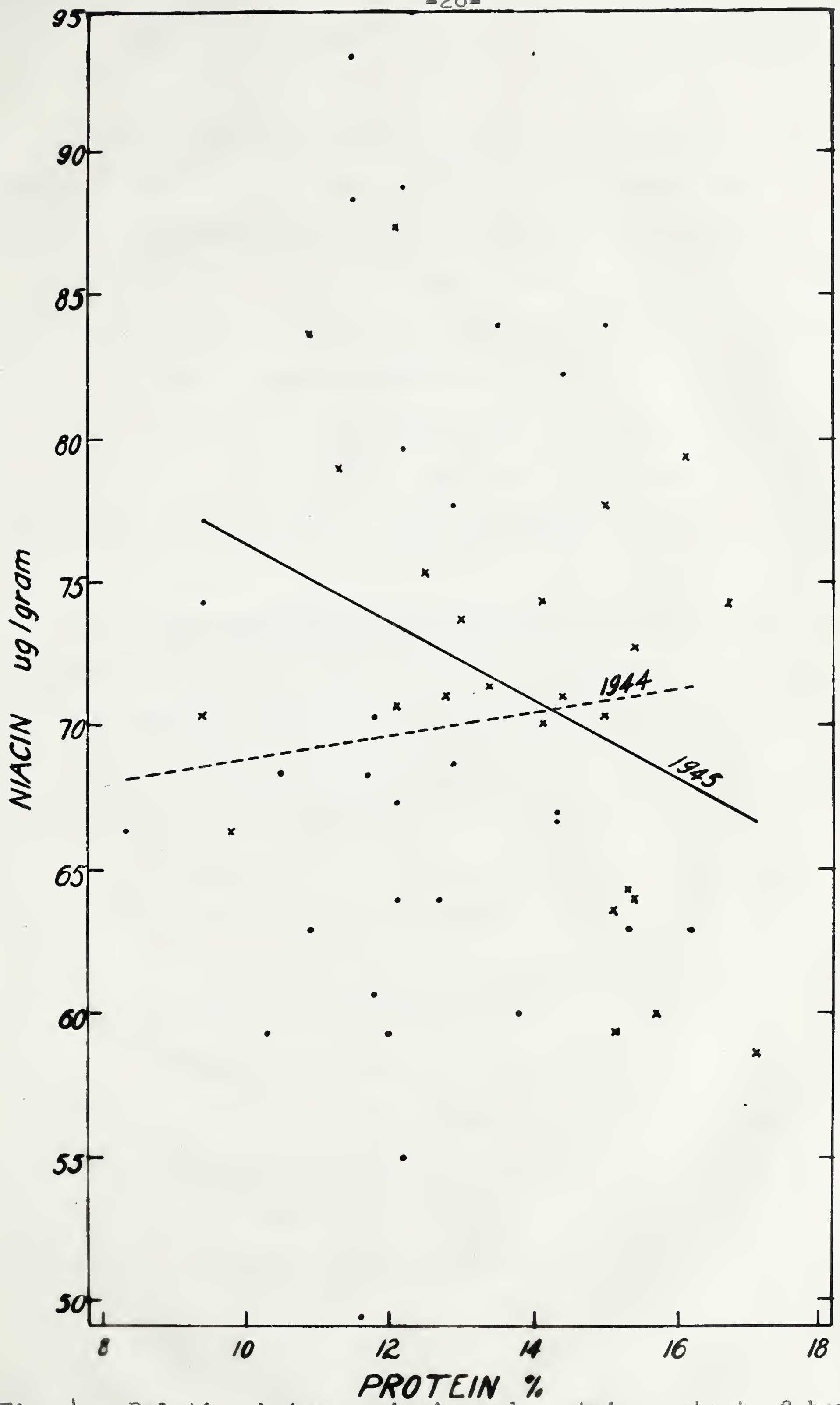

Fig. 4. Relation between niacin and protein content of barley.
1944 ... 1945 xxx

In the later stages of this study an attempt was made to separate and evaluate some of the possible causal agents affecting niacin accumulation. The material used consisted of a number of selections from a soft wheat cross, the parents of which were Bunyip and Dicklow. The hybrid seed was grown in bulk plots until the F4 generation in 1940. At that time random selections were made. Yield tests were carried out at Edmonton and Fallis during 1945 and 1946 using seed from the F7 selections grown in 1944. Each selection was grown in four replicates.

In 1945 the seed from each replicate for each selection was bagged separately and the niacin determinations carried out on the seed from one replicate, chosen at random, from each of twenty-five selections grown at Edmonton and Fallis. In 1946 the seed from the four replicates of each selection was bulked, and niacin determinations conducted on the bulked samples of seed using the same selections chosen in 1945.

Since the selections were of the F8 generation they may possibly be considered as distinct strains. If this be so, the niacin values actually represent the niacin content of twenty-five different strains of wheat grown on two different soil types, black at Edmonton and gray at Fallis, during 1945 and 1946. The results obtained are summarized in Table X. Individual assay values appear in Appendix Tables XIV and XV.

TABLE X

Niacin and Protein Values for Hybrid Wheat Samples Grown at Edmonton and Fallis During 1945 and 1946.

Year	No. of Samples	Niacin Mean µg/g.	Niacin Range µg/g.	Protein Mean %	Protein Range %
		Edmonton			
1945	25	43.6	38.9-52.4	13.0	11.2-15.2
1946	25	50.2	42.2-60.3	10.7	8.8-12.9
Total Mean	50	46.9		11.9	
		Fallis			
1945	25	45.8	39.3-52.9	11.9	10.3-13.2
1946	25	44.3	40.3-49.6	7.2	6.2- 9.0
Total Mean	50	45.1		9.5	

The niacin values obtained on the Edmonton samples averaged 43.6 and 50.2 micrograms per gram for the years 1945 and 1946 respectively with a two year average of 46.9 micrograms per gram. The average values obtained on the Fallis samples were 45.8 and 44.3 micrograms per gram for 1945 and 1946 with a mean of 45.1 micrograms per gram for the two years.

Though there was a highly significant difference between the mean niacin content of the Edmonton samples for 1945 and 1946, such a difference was not found in comparable samples from the same selections grown at Fallis in the same years. Since significant differences between years were obtained for samples of the same strains grown on the same soil type at Edmonton,

it might be postulated that rainfall and other climatic factors, rather than soil and genetic conditions, are the controlling factors in niacin accumulation. However, the data indicate that the niacin content of the 1946 Edmonton samples is significantly different from that of the 1946 Fallis samples. These samples were grown under different weather and soil conditions, and consequently the variability may possibly be attributable, in part, to these environmental factors. The data are definitely limited and the reasons for the apparent differences remain obscure.

To determine whether or not any relationship existed between the niacin and protein content of the samples assayed, correlation coefficients were determined. The data are tabulated in Table XI. Correlation surfaces, depicting the scatter of points obtained by plotting niacin against protein, are shown in Figures 5 and 6.

TABLE XI

Correlation Coefficients Between Protein and Niacin Content of Hybrid Soft Wheat Samples Grown at Edmonton and Fallis During 1945 and 1946

Station	Year	Number of Samples	Correlation Coefficients
Edmonton	1945	25	-0.055
	1946	25	-0.341
Fallis	1945	25	-0.233
	1946	25	-0.080

The data suggest that no significant relationship exists between the niacin and protein content for the soft wheat samples grown at Edmonton and Fallis during 1945 and 1946. It would appear that under similar environmental conditions, niacin accumulation is not related to protein content.

In Table IX it was shown that the relationships between niacin and protein in hard spring wheat were significantly positive in the 1945 and 1946 samples obtained from widely scattered locations in the province. Although protein and niacin do not appear to be associated, it may be that certain environmental conditions conducive to high protein content are also effective in promoting niacin accumulation. The results reported in Tables X and XI were obtained on samples of soft wheat parentage and the observed differences may not necessarily be applicable to hard wheat varieties.

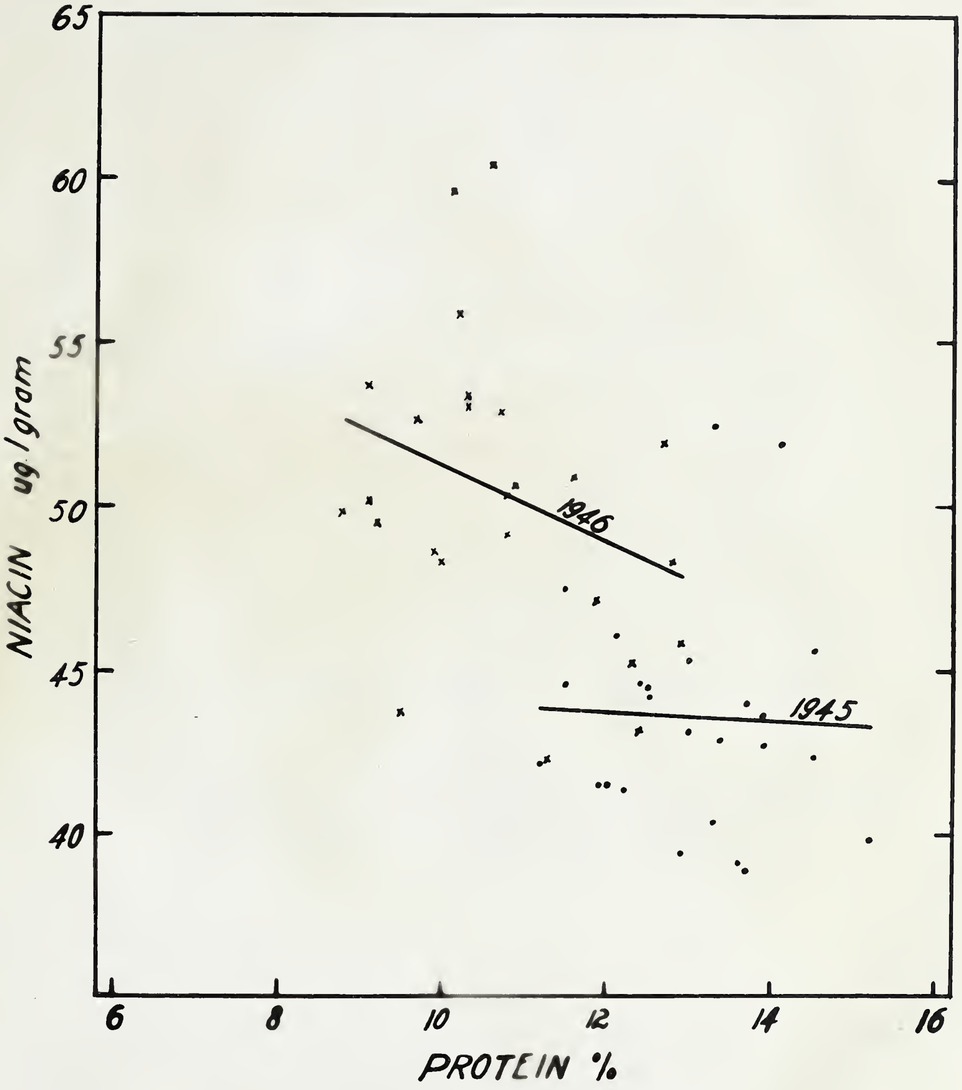

Fig. 5. Relation between niacin and protein content of samples of hybrid wheat grown at Edmonton.
1945 ... 1946 xxx

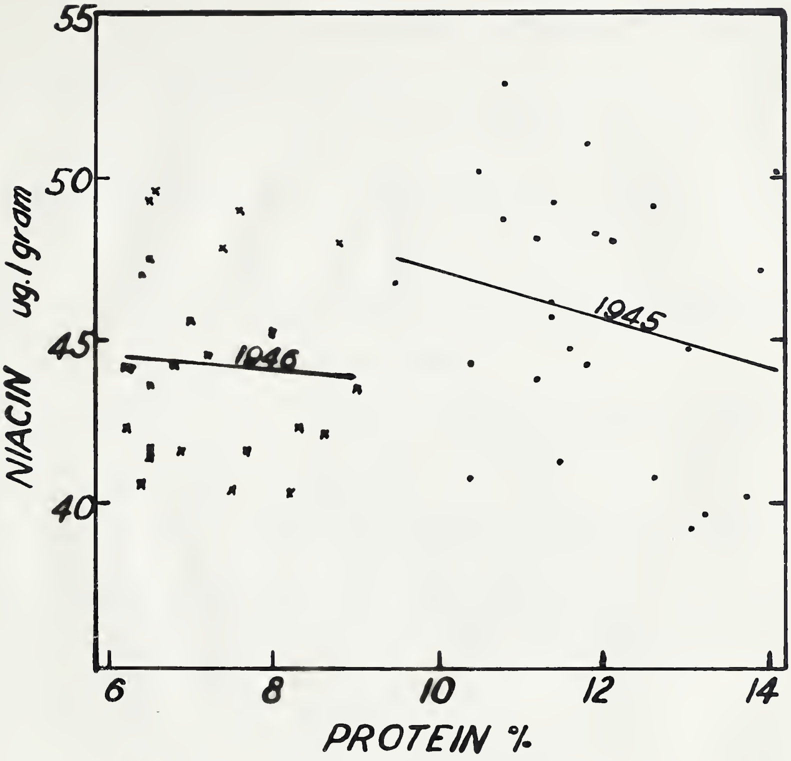

Fig. 6. Relation between niacin and protein content of hybrid wheat grown at Fallis.
1945 ... 1946 xxx

SUMMARY

1. Microbiological niacin assays on wheat, barley and oats have been described.
2. Oats was found to contain the least, hard spring wheat an intermediate and barley the greatest amount of niacin with means of 10.2, 55.4 and 70.7 micrograms per gram respectively.
3. The mean niacin content of the soft wheat strains assayed was 46.0 micrograms per gram.
4. Differences in the niacin content of cereal grains grown in different soil zones and in different years were observed but due to the high variability in niacin content of grains grown in any one soil zone in any one year it was not possible to demonstrate that a real difference, attributable to either soil or year, exists.
5. Assay of samples from widely scattered locations in the province indicated significant positive relationships between the niacin and protein content of 1945 and 1946 wheat and oats. No significant relationships were observed in the barley samples.
6. No significant correlation was observed between the niacin and protein content of 25 selections from the F8 generation of a soft wheat cross grown on both gray and black soil in 1945 and in 1946.

PART I

B. CHEMICAL ASSAYS FOR NIACIN

The assay values reported in the preceding section were obtained microbiologically. In the initial phases of this investigation of the niacin content of Alberta grains attempts were made to assay grains by chemical methods. After preliminary consideration the method of Melnick, Oser and Siegel (25) was adopted for further study. Initially the reproducibility of the results obtained by this method was poor and after considerable experience it was found that a large part of the variability was due to impurities in batches of cyanogen bromide reagent prepared in the laboratory from potassium cyanide and saturated bromine water. When a 0.25 M solution of crystalline cyanogen bromide1) was used the assay results were much more consistent and in better agreement with those obtained microbiologically as well as with values for similar products reported in the literature. Typical values obtained on wheat and oats by the microbiological method and by the chemical method using crystalline cyanogen bromide in one case, and a solution prepared in the laboratory from potassium cyanide and bromine water in the other, are shown in Table XII. The high values obtained when cyanogen bromide prepared in the laboratory was used suggest that this reagent contained interfering elements conducive to increased photometric density in the assay solution.

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1) Purchased from Eastman Kodak Company, Rochester, N.Y.

Repeated attempts to prepare satisfactory solutions of cyanogen bromide from potassium cyanide and bromine water were not successful.

TABLE XII

Comparison of Niacin Values Obtained
Microbiologically and Chemically
(μg. per gram)

Sample	Protein %	Assay Procedure		
		Microbiological	Chemical	
W - 1	19.7	63.0	65.9*	71.7**
2	13.7	55.0	55.0	78.5
3	9.1	65.0	64.0	78.4
O - 1	16.3	20.0	23.6	51.2
2	12.3	9.0	12.0	33.9
3	9.6	10.0	13.5	35.5

* Cyanogen bromide reagent prepared from crystals.
**Cyanogen bromide reagent prepared in the laboratory.

After it was found that reproducible chemical assay values could be obtained when purchased crystalline cyanogen bromide was employed, additional chemical assays were done on the 1945 wheat, oat and barley samples described in Appendix Tables IV - VI. Since these samples had been assayed microbiologically, they afforded excellent material for a comparison of chemical and microbiological assay values. The summarized data are given in Table XIII. The individual assay results appear in Appendix Table XVI.

TABLE XIII

Chemical and Microbiological Niacin Values Obtained on the 1945 Wheat, Oat and Barley Samples.

				Procedure			
				Chemical		Microbiological	
	Number of	Protein %		Niacin μg/g.		Niacin μg/g.	
Cereal	Samples	Mean	Range	Mean	Range	Mean	Range
Wheat	23	14.0	8.7-19.7	60.8	50.2-78.1	59.6	45.7-72.3
Oats	28	12.7	8.1-16.9	13.5	9.5-22.6	12.1	8.0-18.7
Barley	24	13.8	9.4-17.1	62.1	48.9-74.3	71.2	58.7-87.3

The mean values obtained for wheat and oats by the two methods compare very favorably. However, the microbiological procedure gave consistently higher results for the barley samples assayed. While there is no definite explanation for this inconsistency it may be due to:

(a) the presence of some unknown niacin derivative which stimulates microbial growth and is not measured by the cyanogen bromide reaction.

(b) the presence of some constituent other than niacin or its derivatives which has a stimulatory effect on microbial growth.

A survey of the literature discloses that numerous discrepancies in niacin assay values are reported depending upon the material analyzed, the method used and the treatment given the material prior to assay. Brown and coworkers (16, 26, 27, 28) have stressed the fact that cereal extracts contain at least two types of compounds which, by the usual chemical and microbiological assay methods, are measured as niacin. These workers

have suggested selective solvent extraction, and/or treatment with hydrogen peroxide as methods for the removal of compounds which interfere with the determination of nicotinic acid in cereal grain extracts by colorimetric and microbiological procedures. Data are presented to show that, while oxidation with hydrogen peroxide materially lowers the apparent niacin content of some cereal extracts, it results in much better agreement between chemical and microbiological assay values. It is also shown that pure niacin added to cereal extracts is not destroyed by the peroxide treatment.

Niacin assay values obtained by the microbiological and chemical methods previously described were therefore compared with those obtained by a modification of the chemical method outlined by Steele (29) and those obtained by the microbiological method of Snell and Wright on an extract previously treated with Lloyd's reagent and hydrogen peroxide. The values obtained on three samples of barley are shown in Table XIV. The figures given in the last two columns of this table were obtained by the following procedure:

A two gram sample was weighed and suspended in 75 ml. of 2 N H_2SO_4 in a 100 ml. volumetric flask. After hydrolysis in boiling water for one hour the flask was cooled and made up to volume with distilled water. The contents were well mixed and two 25 ml. aliquots, one for chemical and one for microbiological assay, were transferred to 50 ml. centrifuge tubes. Adsorption on Lloyd's reagent, elution with NaOH and decolorization with $Pb(NO_3)_2$ were carried out according to the method of Steele (29).

The niacin content of the extract from the first tube was determined by the color development procedure of Melnick, Oser and Siegel (25). Prior to microbiological assay the extract from the second tube was treated with 5 ml. of 40 per cent NaOH and 5 ml. of 30 per cent hydrogen peroxide at a temperature of 70°C. for two hours and then evaporated to dryness on a steam bath. The residue was dissolved in distilled water, adjusted to pH 6.8 and diluted to the original volume. The oxidized extract was assayed by the microbiological method of Snell and Wright (23).

TABLE XIV

Niacin Assay Values Obtained by Different Methods.

Sample	(1) Microbiological, Snell and Wright	(2) Chemical, Melnick, Oser and Siegel	(3) Chemical following treatment with Lloyd's reagent and $Pb(NO_3)_2$	(4) Microbiological, following treatment with Lloyd's reagent, $Pb(NO_3)_2$ and H_2O_2
	micrograms per gram			
B - 1	63	52	34	36
2	77	66	40	45
3	75	68	43	48

The limited data shown in the above table indicate that barley extracts contain chromogenic substances which are removed by treatment with Lloyd's reagent and $Pb(NO_3)_2$ but are not removed and appear as niacin in the method of Melnick, Oser and Siegel. They also show that niacin assay values obtained microbiologically on extracts treated with Lloyd's reagent, $Pb(NO_3)_2$ and H_2O_2 are are much lower than those secured by the method of Snell and

Wright. These results are in general agreement with those of Brown, Thomas and Bina (16). There is a need for bio-assay results with which the values obtained by chemical and microbiological methods may be compared.

The main purpose of this investigation was to acquire information on the relative niacin content of different cereal grains and on variations that might occur in one cereal grown on different types of soil. In view of this, study of different methods of niacin assay was discontinued, and the microbiological method of Snell and Wright (23) as modified by Krehl, Strong and Elvehjem (24) was adopted for routine assays. This method is essentially the same as the tentative method listed in Methods of Analysis - A.O.A.C. - sixth edition, 1945.

PART II

THE NIACIN CONTENT OF A COMMERCIAL TURKEY STARTER AND COMMENTS ON THE POSSIBILITY OF A NIACIN DEFICIENCY IN POULTRY AND SWINE RATIONS

The importance of niacin in animal and poultry nutrition has been mentioned. While tentative requirements for these species are reported, most of the recommendations are based on experimental results obtained with animals maintained on synthetic diets and may not indicate the requirements under practical conditions. Briggs (10) has indicated that the requirement of the poult is 3 to 5 mg. per 100 grams of ration, a figure 2 to 3 times as great as that generally recommended for chicks (30). In order to insure that this requirement is met, commercial feed companies are beginning to fortify turkey starters with niacin.

The fortified starter assayed in this study is prepared and sold by a local feed company. Samples of the complete starter and the individual components were obtained from the company and were assayed within a few days, using the microbiological procedure previously described. The formula of the starter is given in Table XV. Premix 7 constituted the medium for niacin fortification and consisted of finely ground limestone grit plus niacin.

TABLE XV

Turkey Starter Formula

Component	Lbs. per ton
Ground wheat	384
Ground oats	200
Ground groats	100
Ground barley	260
Shorts	40
Feed flour	16
Wheat bran	100
Alfalfa meal	200
Herring meal	250
Meat meal	300
Buttermilk powder	100
Premix 7	16
Iodized salt	20
Fish oil (400D, 3000A)	10

The weights of the samples and the dry matter equivalent of the hydrolysate aliquot assayed are given in Table XVI.

TABLE XVI

Weight of Sample and Aliquot Assayed

Component	Wt. of Sample (grams)	Aliquot (grams)
Complete starter	1.5	0.0006
Ground wheat	1.5	0.0006
Ground oats	2.0	0.002
Ground groats	2.0	0.002
Ground barley	1.5	0.0006
Shorts	1.0	0.0002
Feed flour	2.0	0.002
Wheat bran	1.0	0.0001
Alfalfa meal	2.0	0.0008
Herring meal	1.0	0.0004
Meat meal	1.5	0.0006
Buttermilk powder	2.0	0.005
Premix 7	2.0	0.00004

The niacin content of the complete starter and the individual components are tabulated in Table XVII.

TABLE XVII

Niacin Content of Mixed Turkey Starter and Individual Components

Component Assayed	µg/g.	mg/100 gms.	mg/lb.	Mixing formula (1000 lbs.)	Niacin Contribution in complete starter mg/100 grams
Complete starter	61	6.1	27.7		6.1
Ground wheat	59	5.9	26.8	192	1.1
Ground oats	10	1.0	4.5	100	0.1
Ground groats	10	1.0	4.5	50	0.1
Ground barley	67	6.7	30.4	130	0.9
Shorts	227	22.7	103.1	20	0.4
Feed flour	24	2.4	10.9	8	0.0
Wheat bran	192	19.2	87.2	50	1.0
Alfalfa meal	24	2.4	10.9	100	0.2
Herring meal	44	4.4	20.0	125	0.6
Meat meal	44	4.4	20.0	150	0.7
Buttermilk powder	6	0.6	2.7	50	0.0
Premix 7	1250	125.0	567.5	8	1.0
Iodized salt	-	-	-	10	
Fish Oil(400D,3000A)	-	-	-	5	
					6.1

On the whole, the values obtained on the various components are in good agreement with the results previously obtained and with values reported in the literature. The niacin content of the shorts sample analyzed in this study is considerably above that reported for this by-product in the literature. This may possibly be accounted for by the fact that the sample of shorts assayed contained a high proportion of bran. Although the niacin value for bran shown in Table XVII is only 192 micrograms per

gram as compared to 227 for shorts, values of 157 to 409 micrograms of niacin per gram have been reported for bran and it is possible that the bran present in this particular sample of shorts may have been relatively rich in niacin.

Briggs (10) has suggested that the niacin requirement of the poult may be on the order of 3 to 5 milligrams per 100 grams of ration. Analysis of the complete starter used in the present study indicated a niacin content of 6.1 milligrams per 100 grams of starter. The niacin content obtained by the additive contribution of the various components also gave a value of 6.1 milligrams per 100 grams. If the niacin contributed through premix 7 were omitted, the niacin content of the starter would be 5.1 milligrams per 100 grams, which would still be enough to meet the recommendations of Briggs.

If it is assumed that the niacin values obtained by microbiological assay represent the true niacin value for turkeys and if it is further assumed that 3 to 5 milligrams of niacin per 100 grams of practical starter is adequate for the turkey poult, it would not appear necessary to fortify this starter with premix 7. Practical feeding trials are required to settle the question of the necessity of fortifying turkey starters with niacin.

The niacin requirement of chicks is believed to be considerably lower than that of poults and Briggs *et al.* (11) have suggested that there is little possibility of a niacin deficiency in practical chick rations.

Hughes (12), on the basis of results obtained with a purified ration, has reported the niacin requirement of pigs as 5 to 10 mg. per 100 pounds of live weight. In a report prepared by the committee on animal nutrition of the National Research Council (31) the requirement is given as 12.5 mg. per 100 pounds of live weight. Wheat, oats and barley comprise the main constituents of swine rations in Alberta. According to the values obtained in the present study, wheat contains approximately 25, oats 4 to 5 and barley 32 mg. of niacin per pound. Assuming that a hog normally consumes about 4% of its weight in feed per day, a total daily niacin consumption of 18.0 mg. would be obtained if oats constituted the only cereal in the ration. In Western Canada, swine rations are, in the main, made up from mixtures of oats and barley, and since barley is a relatively good source of the vitamin, the possibility of a niacin deficiency in such rations is small. This conclusion is substantiated by the results of experiments conducted at the University of Alberta (32) in which it was found that fortification of growing pig rations with niacin failed to promote any improvement in rate of gain or economy of use of feed.

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APPENDIX TABLES

Appendix Table 1

Protein and Niacin Content of Wheat Samples From the Various Soil Zones of Alberta, 1944.

Station	Soil Zone*	Sample No.	Variety	Protein** %	Niacin** µg/g.
Athabasca	G.T.	4 - W - 9	Marquis	8.6	46.3
Fallis	G.	2	Red Bobs	8.9	35.3
Goodfare	G.	24	Marquis	8.9	43.7
Claresholm	D.B.	14	Red Bobs	10.6	38.7
Winfield	G.	8	Marquis	10.6	49.3
Fallis	G.	12	"	10.7	53.7
Debolt	G.T.	22	"	10.9	47.0
Bon Accord	B.	10	"	12.4	74.0
Stettler	S.B.	6	"	12.6	54.7
Falher	G.T.	23	"	13.0	43.7
Craigmyle	D.B.	15	"	13.1	41.3
St. Paul	B.	7	"	13.2	56.7
Castor	D.B.	4	"	13.5	42.3
Baldonnel	G.	20	"	13.6	44.3
Nobleford	D.B.	18	"	13.7	49.7
Pincher Creek	S.B.	19	Red Bobs	14.2	62.3
Lomond	Br.	17	Marquis	14.2	51.7
Edmonton	B.	11	"	14.4	57.3
Consort	Br.	5	"	14.4	43.0
Airdrie	S.B.	3	"	15.1	54.0
Beaverlodge	B.T.	21	"	15.2	57.3
Drumheller	D.B.	16	"	15.3	53.0
Acadia Valley	Br.	13	"	15.4	52.0
Conrad	Br.	1	?	17.3	44.7
			Total	309.8	1196.0
			Number of Samples	24	24
			Mean	12.9	49.8
			Min.	8.6	35.3
			Max.	17.3	74.0

*In this and subsequent tables: G.=Gray wooded; G.T.=Gray transitional; B.=Black; B.T.=Black transitional; S.B.=Shallow black; D.B.=Dark brown; Br.=Brown.

**Basis 13.5% moisture in all tables.

Appendix Table II

Protein and Niacin Content of Oat Samples from the Various Soil Zones of Alberta, 1944.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
Goodfare	G.	4 - 0 - 27	Victory	6.6	7.3
Bon Accord	G.T.	3	"	8.2	14.3
Chedderville	G.	7	"	8.3	7.0
Baldonnel	G.	23	"	8.3	7.0
Winfield	G.	13	"	8.8	7.0
Claresholm	D.B.	17	"	8.9	7.7
Fallis	G.	15	"	9.3	7.0
Bon Accord	B.	14	"	9.5	7.3
Lomond	Br.	20	"	9.5	11.3
Dickson	B.T.	9	"	10.8	7.0
Beaverlodge	B.T.	24	"	10.9	9.7
Stettler	S.B.	11	"	11.0	7.0
Craigmyle	D.B.	18	"	11.2	9.3
Falher	G.T.	26	"	11.3	7.3
Edmonton	B.	16	"	11.4	8.7
Consort	Br.	8	"	11.5	8.0
Nobleford	D.B.	21	"	11.6	10.3
Fallis	G.	10	"	11.9	8.0
Airdrie	S.B.	4	"	12.2	9.0
Castor	D.B.	5	"	12.3	8.0
Debolt	G.T.	25	"	12.3	10.3
Edmonton	B.	2	"	12.7	9.0
St. Paul	B.	12	"	13.1	8.3
Chauvin	D.B.	6	"	13.4	8.3
Pincher Creek	S.B.	22	"	13.6	9.3
Drumheller	D.B.	19	Eagle	13.7	9.3
Wrentham	Br.	1	Victory	15.9	13.0
		Total		298.2	235.7
		Number of Samples		27	27
		Mean		11.0	8.7
		Min.		6.6	7.0
		Max.		15.9	14.3

Appendix Table III

Protein and Niacin Content of Barley Samples from the Various Soil Zones of Alberta, 1944.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin ug/g.
Goodfare	G.	4 - B - 29	Newal	8.3	66.3
Bon Accord	G.T.	3	?	9.4	74.3
Claresholm	D.B.	19	Newal	10.3	59.3
Debolt	G.T.	27	"	10.5	68.3
Acadia Valley	Br.	18	"	10.9	63.0
Chedderville	G.	7	"	11.5	87.3
Dickson	B.T.	9	"	11.5	93.3
Nobleford	D.B.	23	"	11.6	49.3
Baldonnel	G.	25	"	11.7	68.3
Beaverlodge	B.T.	26	"	11.8	70.3
Stettler	S.B.	12	"	11.8	60.7
Falher	G.T.	28	"	12.0	59.3
Lomond	Br.	22	"	12.1	64.0
Castor	D.B.	5	"	12.1	67.3
Airdrie	S.B.	4	"	12.2	88.7
Fallis	G.	17	"	12.2	79.7
Drumheller	D.B.	20	"	12.2	55.0
Clover Bar	B.	2	"	12.7	64.0
Consort	Br.	8	"	12.9	68.7
Bon Accord	B.	15	"	12.9	77.7
Leslieville	B.T.	10	"	13.5	84.0
Craigmyle	D.B.	21	"	13.8	60.0
Edmonton	B.	16	"	14.3	66.7
St. Paul	B.	13	"	14.3	67.0
Chauvin	D.B.	6	"	14.4	82.3
Metiskow	D.B.	11	"	15.0	84.0
Pincher Creek	S.B.	24	"	15.3	63.0
Conrad	Br.	1	Trebi	16.2	63.0
			Total	347.4	1954.8
			Number of Samples	28	28
			Mean	12.4	69.8
			Min.	8.3	49.3
			Max.	16.2	93.3

Appendix Table IV

Protein and Niacin Content of Wheat Samples from Various Soil Zones of Alberta, 1945.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
Metiskow	D.B.	5 - W - 9	Marquis	8.7	52.0
Fallis	G.	3	"	9.2	49.3
Goodfare	G.	23	Thatcher	11.3	45.7
Chauvin	D.B.	6	Marquis	12.0	53.7
Acadia Valley	Br.	12	"	12.5	58.3
Baldonnel	G.	18	Thatcher	12.5	66.3
Beaverlodge	B.T.	19	"	12.7	69.3
Airdrie	D.B.	4	Marquis	12.7	62.0
Bremner	B.	2	"	13.7	53.0
Debolt	G.T.	20	Thatcher	13.8	64.0
Pincher Creek	D.B.	13	Red Bobs	14.0	61.7
St. Paul	B.	5	Marquis	14.1	49.3
Nobleford	D.B.	17	"	14.5	66.7
Athabasca	G.T.	25	"	14.8	59.3
Falher	G.T.	22	Thatcher	15.0	52.7
Drumheller	D.B.	8	Marquis	15.3	54.7
Castor	D.B.	7	"	15.4	54.7
Craigmyle	D.B.	15		15.5	60.3
Dickson	B.T.	10	Marquis	15.8	60.3
Lomond	Br.	16	Thatcher	15.9	72.3
Fairview	G.	21	"	16.0	72.3
Drumheller	D.B.	14	Marquis	16.4	66.7
Conrad	Br.	1	"	19.7	66.7
			Total	321.5	1371.3
			Number of Samples	23	23
			Mean	14.0	59.6
			Min.	8.7	45.7
			Max.	19.7	72.3

Appendix Table V

Protein and Niacin Content of Oat Samples From the Various Soil Zones of Alberta, 1945.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
Chedderville	G.	5 - 0 - 10	Victory	8.1	8.0
Baldonnel	G.	19	"	9.1	10.7
Warburg	G.	3	Banner	9.6	9.7
Beaverlodge	B.T.	20	Victory	9.8	14.3
Goodfare	G.	24	"	10.8	11.3
Airdrie	D.B.	4	"	10.9	8.7
Fairview	G.	22	"	11.7	11.0
Fallis	G.	12	"	11.5	13.7
Debolt	G.T.	21	"	11.9	12.7
Progress	G.T.	25	"	12.0	10.0
Clover Bar	B.	2	Banner	12.3	8.3
Dickson	B.T.	11	Victory	12.3	11.3
Athabasca	G.T.	26	"	12.4	16.7
Pincher Creek	D.B.	14	"	12.8	10.7
Warburg	G.	28	"	12.7	13.3
Metiskow	D.B.	9	"	12.8	9.0
Bon Accord	B.	27	"	13.1	14.3
Castor	D.B.	7	"	13.4	9.7
Chauvin	D.B.	6	"	13.6	9.0
Drumheller	D.B.	8	"	13.8	15.0
St. Paul	B.	5	"	13.7	10.3
Falher	G.T.	23	"	14.3	12.3
Drumheller	D.B.	15	"	14.3	13.7
Nobleford	D.B.	18	"	14.5	12.0
Craigmyle	D.B.	16	"	14.9	12.3
Taber	Br.	1	Banner	16.3	18.7
Lomond	Br.	17	Ajax	16.5	14.7
Acadia Valley	Br.	13	Victory	16.9	17.0
			Total	356.0	338.4
			Number of Samples	28	28
			Mean	12.7	12.1
			Min.	8.1	8.0
			Max.	16.9	18.7

Appendix Table VI

Protein and Niacin Content of Barley Samples From the Various Soil Zones of Alberta, 1945.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
Stony Plain	B.	5 - B - 3	Olli	9.4	70.3
Chedderville	G.	10	Newal	9.8	66.3
Baldonnel	G.	19	"	10.9	83.7
Debolt	G.T.	21	"	11.3	79.0
Airdrie	D.B.	4	"	12.1	87.3
Fallis	G.	12	"	12.1	70.7
Progress	G.T.	25	"	12.5	75.3
Fairview	G.	22	"	12.8	71.0
Mundare	B.	2	Olli	13.0	73.7
Dickson	B.T.	11	Newal	13.4	71.3
Metiskow	D.B.	9	"	14.1	70.0
Lomond	Br.	17	"	14.1	74.3
Acadia Valley	Br.	13	"	14.4	71.0
Pincher Creek	D.B.	14	"	15.0	77.7
Drumheller	D.B.	15	"	15.0	70.3
St. Paul	B.	5	"	15.1	63.7
Drumheller	D.B.	8	"	15.1	59.3
Chauvin	D.B.	6	"	15.3	64.3
Craigmyle	D.B.	16	"	15.4	72.7
Falher	G.T.	23	"	15.4	64.0
Castor	D.B.	7	"	15.7	60.0
Beaverlodge	B.T.	20	"	16.1	79.3
Nobleford	D.B.	18	"	16.7	74.3
Legend	Br.	1	"	17.1	58.7
			Total	331.8	1708.2
			Number of Samples	24	24
			Mean	13.8	71.2
			Min.	9.4	58.7
			Max.	17.1	87.3

Appendix Table VII

Protein and Niacin Content of Wheat Samples from the Various Soil Zones of Alberta, 1946.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
Chauvin	D.B.	6 - W - 1	Thatcher	16.9	77.9
St. Albert	G.T.	2	Red Bobs	11.6	52.7
Villeneuve	G.	3	Garnet	8.7	43.4
Metiskow	D.B.	4	Thatcher	14.5	56.2
St. Paul	B.	5	"	12.9	61.5
Castor	D.B.	6	"	14.6	61.0
Consort	D.B.	7	"	14.3	69.8
Dickson	B.T.	8	"	10.8	56.4
			Total	104.3	478.9
		Number	of Samples	8	8
			Mean	13.0	59.9
			Min.	8.7	43.4
			Max.	16.9	77.9

Appendix Table VIII

Protein and Niacin Content of Barley Samples from the Various Soil Zones of Alberta, 1946.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
Wheat Center	B.	6 - B - 1	-	12.9	57.9
Legal	G.T.	2	-	12.7	76.6
Morinville	G.T.	3	Olli	9.4	70.9
Metiskow	D.B.	4	Newal	14.8	71.2
St. Paul	B.	5	"	14.3	73.7
Castor	D.B.	6	"	14.8	71.6
Consort	D.B.	7	"	15.9	69.1
Dickson	B.T.	8	"	11.6	85.4
			Total	106.4	576.4
			Number of Samples	8	8
			Mean	13.3	72.0
			Min.	9.4	57.9
			Max.	15.9	85.4

Appendix Table IX

Protein and Niacin Content of Oat Samples from the Various Soil Zones of Alberta, 1946.

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
New Dayton	B.	6 - 0 - 1	-	14.9	11.6
St. Albert	G.T.	2	Victory	13.1	8.8
Bon Accord	G.	3	-	10.3	7.3
Metiskow	D.B.	4	Victory	13.0	8.2
St. Paul	B.	5	"	12.4	8.9
Castor	D.B.	6	"	13.1	8.5
Consort	D.B.	7	"	13.2	8.9
Dickson	B.T.	8	"	10.7	8.6
			Total	100.7	70.8
			Number of Samples	8	8
			Mean	12.6	8.8
			Min.	10.3	7.3
			Max.	14.9	11.6

Appendix Table X

Protein and Niacin Content of 1944 and 1945 **Marquis** Wheat Samples Grouped According to Soil Zone

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin μg/g.
1944					
Athabasca	G. & G.T.	4 - W - 9	Marquis	8.6	46.3
Goodfare	"	24	"	8.9	43.7
Winfield	"	8	"	10.6	49.3
Fallis	"	12	"	10.7	53.7
Debolt	"	22	"	10.9	47.0
Falher	"	23	"	13.0	43.7
Baldonnel	"	20	"	13.6	44.3
			Mean	10.9	46.9
1945					
Fallis					
Athabasca	"	5 - W - 3	"	9.2	49.3
		25	"	14.8	59.3
			Mean	12.0	54.3
1944					
Bon Accord	B., S.B. & B.T.	4 - W -10	"	12.4	74.0
Stettler	"	6	"	12.6	54.7
St. Paul	"	7	"	13.2	56.7
Edmonton	"	11	"	14.4	57.3
Airdrie	"	3	"	15.1	54.0
Beaverlodge	"	21	"	15.2	57.3
			Mean	13.8	59.0
1945					
Bremner	"	5 - W - 2	"	13.7	53.0
St. Paul	"	5	"	14.1	49.3
Dickson	"	10	"	15.8	60.3
			Mean	14.5	54.2

Appendix Table X (continued)

Station	Soil Zone	Sample No.	Variety		Protein %	Niacin ug/g.
1944						
Craigmyle	Br. & D.B.	4 - W - 15	Marquis		13.1	41.3
Castor	"	4	"		13.5	42.3
Nobleford	"	18	"		13.7	49.7
Drumheller	"	16	"		15.3	53.0
Lomond	"	17	"		14.2	51.7
Consort	"	5	"		14.4	43.0
Acadia Valley	"	13	"		15.4	52.0
				Mean	14.2	47.6
1945						
Metiskow	"	5 - W - 9	"		8.7	52.0
Chauvin	"	6	"		12.0	53.7
Airdrie	"	4	"		12.7	62.0
Nobleford	"	17	"		14.5	66.7
Drumheller	"	8	"		15.3	54.7
Castor	"	7	"		15.4	54.7
Drumheller	"	14	"		16.4	66.7
Acadia Valley	"	12	"		12.5	58.3
Conrad	"	1	"		19.7	66.7
				Mean	14.1	59.5

Appendix Table XI

Protein and Niacin Content of 1944 and 1945 Victory Oat Samples Grouped According to Soil Zone

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin μg/g.
1944					
Goodfare	G. & G.T.	4 - 0 - 27	Victory	6.6	7.3
Bon Accord	"	3	"	8.2	14.3
Chedderville	"	7	"	8.3	7.0
Baldonnel	"	23	"	8.3	7.0
Winfield	"	13	"	8.8	7.0
Fallis	"	15	"	9.3	7.0
Falher	"	26	"	11.3	7.3
Fallis	"	10	"	11.9	8.0
Debolt	"	25	"	12.3	10.3
			Mean	9.4	8.4
1945					
Chedderville	"	5 - 0 - 10	"	8.1	8.0
Baldonnel	"	19	"	9.1	10.7
Goodfare	"	24	"	10.8	11.3
Fallis	"	12	"	11.5	13.7
Fairview	"	22	"	11.7	11.0
Debolt	"	21	"	11.9	12.7
Progress	"	25	"	12.0	10.0
Athabasca	"	26	"	12.4	16.7
Warburg	"	28	"	12.7	13.3
Falher	"	23	"	14.3	12.3
			Mean	11.5	12.0

Appendix Table XI (continued)

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
1944					
Bon Accord	B., S.B., & B.T.	4 - 0 - 14	Victory	9.5	7.3
Dickson	"	9	"	10.8	7.0
Beaverlodge	"	24	"	10.9	9.7
Stettler	"	11	"	11.0	7.0
Edmonton	"	16	"	11.4	8.7
Airdrie	"	4	"	12.2	9.0
Edmonton	"	2	"	12.7	9.0
St. Paul	"	12	"	13.1	8.3
Pincher Creek	"	22	"	13.6	9.3
			Mean	11.7	8.4
1945					
Beaverlodge	"	5 - 0 - 20	"	9.8	14.3
Dickson	"	11	"	12.3	11.3
Bon Accord	"	27	"	13.1	14.3
St. Paul	"	5	"	13.7	10.3
			Mean	12.2	12.6
1944					
Claresholm	Br. & D.B.	4 - 0 - 17	"	8.9	7.7
Craigmyle	"	18	"	11.2	9.3
Nobleford	"	21	"	11.6	10.3
Castor	"	5	"	12.3	8.0
Chauvin	"	6	"	13.4	8.3
Lomond	"	20	"	9.5	11.3
Consort	"	8	"	11.5	8.0
Wrentham	"	1	"	15.9	13.0
			Mean	11.8	9.5

Appendix Table XI (continued)

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin ug/g.
1945					
Airdrie	Br. & D.B.	5 - 0 - 4	Victory	10.9	8.7
Metiskow	"	9	"	12.8	9.0
Pincher Creek	"	14	"	12.8	10.7
Castor	"	7	"	13.4	9.7
Chauvin	"	6	"	13.6	9.0
Drumheller	"	8	"	13.8	15.0
Drumheller	"	15	"	14.3	13.7
Nobleford	"	18	"	14.5	12.0
Craigmyle	"	16	"	14.9	12.3
Acadia Valley	"	13	"	16.9	17.0
			Mean	13.8	11.7

Appendix Table XII

Protein and Niacin Content of 1944 and 1945 Newal Barley Samples Grouped According to Soil Zone

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin ug/g.
1944					
Goodfare	G. & G.T.	4 - B - 29	Newal	8.3	66.3
Debolt	"	27	"	10.5	68.3
Chedderville	"	7	"	11.5	87.3
Baldonnel	"	25	"	11.7	68.3
Falher	"	28	"	12.0	59.3
Fallis	"	17	"	12.2	79.7
			Mean	11.0	71.5
1945					
Chedderville	"	5 - B - 10	"	9.8	66.3
Baldonnel	"	19	"	10.9	83.7
Debolt	"	21	"	11.3	79.0
Fallis	"	12	"	12.1	70.7
Progress	"	25	"	12.5	75.3
Fairview	"	22	"	12.8	71.0
Falher	"	23	"	15.4	64.0
			Mean	12.1	72.9
1944					
Dickson	B., S.B. & B.T.	4 - B- 9	"	11.5	93.3
Beaverlodge		26	"	11.8	70.3
Stettler	"	12	"	11.8	60.7
Airdrie	"	4	"	12.2	88.7
Clover Bar	"	2	"	12.7	64.0
Bon Accord	"	15	"	12.9	77.7

Appendix Table XII (continued)

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
1944 (continued)					
Leslieville	B., S.B. & B.T.	4 - B - 10	Newal	13.5	84.0
Edmonton		16	"	14.3	66.7
St. Paul	"	13	"	14.3	67.0
Pincher Creek	"	24	"	15.3	63.0
			Mean	13.0	73.5
1945					
Dickson	"	5 - B - 11	"	13.4	71.3
St. Paul	"	5	"	15.1	63.7
Beaverlodge	"	20	"	16.1	79.3
			Mean	14.9	71.4
1944					
Claresholm	Br. & D.B.	4 - B - 19	"	10.3	59.3
Nobleford	"	23	"	11.6	49.3
Castor	"	5	"	12.1	67.3
Drumheller	"	20	"	12.2	55.0
Craigmyle	"	21	"	13.8	60.0
Chauvin	"	6	"	14.4	82.3
Metiskow	"	11	"	15.0	84.0
Acadia Valley	"	18	"	10.9	63.0
Lomond	"	22	"	12.1	64.0
Consort	"	8	"	12.9	68.7
			Mean	12.5	65.3

Appendix Table XII (continued)

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
1945					
Airdrie	Br. & D.B.	5 - B - 4	Newal	12.1	87.3
Metiskow	"	9	"	14.1	70.0
Pincher Creek	"	14	"	15.0	77.7
Drumheller	"	15	"	15.0	70.3
Drumheller	"	8	"	15.1	59.3
Chauvin	"	6	"	15.3	64.3
Craigmyle	"	16	"	15.4	72.7
Castor	"	7	"	15.7	60.0
Nobleford	"	18	"	16.7	74.3
Legend	"	1	"	17.1	58.7
Lomond	"	17	"	14.1	74.3
Acadia Valley	"	13	"	14.4	71.0
			Mean	15.0	70.0

Appendix Table XIII

Protein and Niacin Content of Miscellaneous 1944 and 1945 Wheat, Oat and Barley Samples

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
(1) Wheat					
1944					
Conrad	Br.	4 - W - 1	-	17.3	44.7
Fallis	G.	2	Red Bobs	8.9	35.3
Claresholm	D.B.	14	" "	10.6	38.7
Pincher Creek	S.B.	19	" "	14.2	62.3
1945					
Pincher Creek	D.B.	5 - W - 13	" "	14.0	61.7
Craigmyle	"	15	" "	15.5	60.3
Lomond	Br.	16	Thatcher	15.9	72.3
Baldonnel	G.	18	"	12.5	66.3
Beaverlodge	B.T.	19	"	12.7	69.3
Debolt	G.T.	20	"	13.8	64.0
Fairview	G.	21	"	16.0	72.3
Falher	G.T.	22	"	15.0	52.7
Goodfare	G.	23	"	11.3	45.7
(2) Oats					
1944					
Drumheller	D.B.	4 - 0 - 19	Eagle	13.7	9.3
1945					
Taber	Br.	5 - 0 - 1	Banner	16.3	18.7
Clover Bar	B.	2	"	12.3	8.3
Warburg	G.	3	"	9.6	9.7
Lomond	Br.	17	Ajax	16.5	14.7

Appendix Table XIII (continued)

Station	Soil Zone	Sample No.	Variety	Protein %	Niacin µg/g.
(3) Barley					
1944					
Conrad	Br.	4 - B - 1	Trebi	16.2	63.0
Bon Accord	G.T.	3	-	9.4	74.3
1945					
Mundare	B.	5 - B - 2	Olli	13.0	73.7
Stony Plain	B.	3	"	9.4	70.3

Appendix Table XIV

Protein and Niacin Content of Hybrid Soft Wheat Samples Grown at Edmonton during 1945 and 1946.

	1945		1946	
Sample No.	Protein %	Niacin µg/g.	Protein %	Niacin µg/g.
H-W-45-02	12.5	44.2	12.7	51.9
05	14.1	51.9	10.6	60.3
08	14.5	45.6	10.3	53.3
17	13.9	42.7	12.4	43.1
18	11.9	41.5	9.1	50.2
24	13.9	43.6	12.8	48.4
28	13.7	44.0	11.9	47.2
30	11.5	47.5	10.9	50.6
31	12.1	46.1	10.1	59.6
32	13.0	45.3	9.7	52.6
47	13.4	42.9	10.8	49.1
48	14.5	42.3	9.2	49.5
51	12.5	44.5	9.9	48.7
55	13.3	40.3	10.3	53.0
56	12.2	41.3	8.8	49.8
68	11.2	42.1	10.2	55.8
79	12.9	39.4	12.3	45.2
80	12.0	41.5	11.3	42.2
82	11.5	44.6	10.0	48.4
84	13.0	43.1	9.1	53.7
86	15.2	39.9	9.5	43.7
88	13.7	38.9	12.9	45.8
89	13.6	39.1	11.6	50.8
91	13.3	52.4	10.7	52.8
98	12.4	44.7	10.8	50.2
Total	325.8	1089.4	267.9	1255.9
No. of Samples	25	25	25	25
Mean	13.0	43.6	10.7	50.2
Min.	11.2	38.9	8.8	42.2
Max.	15.2	52.4	12.9	60.3

Appendix Table XV

Protein and Niacin Content of Hybrid Soft Wheat Samples Grown at Fallis during 1945 and 1946.

Sample No.	1945 Protein %	1945 Niacin μg/g.	1946 Protein %	1946 Niacin μg/g.
H-W-45-02	11.6	44.8	8.8	48.0
05	12.6	49.1	6.5	49.3
08	13.1	39.3	6.4	40.6
17	10.8	52.9	8.3	42.3
18	10.5	50.1	6.2	44.2
24	9.5	46.8	8.0	45.3
28	11.5	41.3	7.6	49.0
30	11.9	48.3	6.9	41.6
31	11.8	51.0	6.2	44.2
32	11.2	43.9	7.4	47.8
47	11.8	44.3	6.6	49.6
48	10.8	48.7	6.8	44.4
51	11.2	48.1	6.5	47.5
55	12.6	40.8	6.5	43.6
56	13.7	40.2	7.0	45.7
68	10.4	40.7	6.5	41.6
79	12.1	48.0	7.7	41.6
80	11.4	45.6	8.6	42.1
82	13.9	47.1	7.2	44.5
84	13.2	39.7	6.2	42.3
86	10.4	44.4	6.4	47.0
88	11.4	46.1	9.0	43.5
89	13.0	44.7	8.2	40.3
91	14.1	50.1	7.5	40.4
98	11.4	49.2	6.5	41.5
Total	295.9	1145.2	179.5	1107.9
No. of Samples	25	25	25	25
Mean	11.8	45.8	7.2	44.3
Min.	9.5	39.3	6.2	40.3
Max.	14.1	52.9	9.0	49.6

Appendix Table XVI

Comparison of Chemical and Microbiological Niacin Assay Values obtained on Wheat, Oats and Barley Grown in 1945.

Wheat				Oats				Barley			
Sample	Protein %	Niacin µg/g. Chem.	Niacin µg/g. Micro.	Sample	Protein %	Niacin µg/g. Chem.	Niacin µg/g. Micro.	Sample	Protein %	Niacin µg/g. Chem.	Niacin µg/g. Micro.
5-W-1	19.7	61.5	66.7	5-O-1	16.3	22.4	18.7	5-B-1	17.1	48.9	58.7
2	13.7	50.2	53.0	2	12.3	11.3	8.3	2	13.0	63.0	73.7
3	9.2	55.3	49.3	3	9.6	12.2	9.7	3	9.4	63.2	70.3
4	12.7	64.2	62.0	4	10.9	12.3	8.7	4	12.1	65.3	87.3
5	14.1	52.0	49.3	5	13.7	11.4	10.3	5	15.1	62.7	63.7
6	12.0	54.7	53.7	6	13.6	10.4	9.0	6	15.3	64.3	64.3
7	15.4	58.0	54.7	7	13.4	10.4	9.7	7	15.7	60.3	60.0
8	15.3	62.3	54.7	8	13.8	15.0	15.0	8	15.1	59.1	59.3
9	8.7	50.7	52.0	9	12.8	9.5	9.0	9	14.0	67.5	70.0
10	15.8	73.0	60.3	10	8.1	9.5	8.0	10	9.8	68.5	66.3
				11	12.3	12.3	11.3	11	13.4	56.5	71.3
12	12.5	64.9	58.3	12	11.5	14.1	13.7	12	12.1	67.1	70.7
13	14.0	65.6	61.7	13	16.9	15.8	17.0	13	14.4	65.3	71.0
14	16.4	65.9	66.7	14	12.8	12.0	10.7	14	15.0	68.9	77.7
15	15.5	63.3	60.3	15	14.3	13.1	13.7	15	15.0	56.6	70.3
16	15.9	78.1	72.3	16	14.9	13.0	12.3	16	15.4	60.5	72.7
17	14.5	57.6	66.7	17	16.5	14.0	14.7	17	14.1	60.0	74.3
18	12.5	62.4	66.3	18	14.5	14.9	12.0	18	16.7	63.0	74.3
19	12.7	66.4	69.3	19	9.1	12.9	10.7	19	10.9	74.3	83.7
20	13.8	60.8	64.0	20	9.8	17.5	14.3	20	16.1	57.1	79.3
21	16.0	67.8	72.3	21	11.9	12.0	12.7	21	11.3	67.0	79.0
22	15.0	52.2	52.7	22	11.7	10.1	11.0	22	12.8	59.2	71.0
23	11.3	57.8	45.7	23	14.3	12.9	12.3	23	15.4	51.9	64.0
				24	10.8	12.0	11.3				
25	14.8	52.8	59.3	25	12.0	11.9	10.0	25	12.5	60.6	75.3
				26	12.4	22.6	16.7				
				27	13.1	16.1	14.3				
				28	12.7	15.1	13.3				

Appendix Table XVI (continued) 2

	Wheat				Oats				Barley		
Sample	Protein %	Niacin µg/g. Chem.	Niacin µg/g. Micro.	Sample	Protein %	Niacin µg/g. Chem.	Niacin µg/g. Micro.	Sample	Protein %	Niacin µg/g. Chem.	Niacin µg/g. Micro.
Total	321.5	1397.5	1371.3		356.0	376.7	338.4		331.7	1490.8	1708.2
No. of Samples	23	23	23		28	28	28		24	24	24
Mean	14.0	60.8	59.6		12.7	13.5	12.1		13.8	62.1	71.2
Min.	8.7	50.2	45.7		8.1	9.5	8.0		9.4	48.9	58.7
Max.	19.7	78.1	72.3		16.9	22.6	18.7		17.1	74.3	87.3

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